

PRESSURE NATRIURESIS IN ISOLATED KIDNEYS
FROM HYPERTENSION PRONE AND HYPERTENSION
RESISTANT RATS (DAHL RATS)

THESE

présentée à la Faculté de médecine de l'Université de Genève
pour obtenir le grade de docteur en médecine

par

Eric GIRARDIN

de

Genève

Thèse No 3587

GENEVE
Editions Médecine et Hygiène
1977

**PRESSURE NATRIURESIS IN ISOLATED KIDNEYS
FROM HYPERTENSION PRONE AND HYPERTENSION
RESISTANT RATS (DAHL RATS)**

THESE

présentée à la Faculté de médecine de l'Université de Genève
pour obtenir le grade de docteur en médecine

par

Eric GIRARDIN

de

Genève

Thèse No 3587

GENEVE
Editions Médecine et Hygiène
1977

La Faculté de médecine, sur le préavis du professeur Alex F. Muller, autorise l'impression de la présente thèse, sans prétendre par là émettre d'opinion sur les propositions qui y sont énoncées.

Genève, le 20 mai 1977

Le doyen :
Marcel Jenny

Thèse No 3587



Je remercie le docteur A. Grandchamp de son aide précieuse et de ses conseils prodigués tout au long de ce travail.

Je remercie le professeur M. Lindheimer pour sa lecture critique du manuscrit.

Je remercie Mesdemoiselles Courtieu et Montendon pour leur collaboration lors du travail expérimental.



Digitized by the Internet Archive
in 2026

https://archive.org/details/IA41545929_0031

INTRODUCTION

The kidney responds to increases in perfusion pressure with increments in sodium and water excretion (1) (2). Guyton et al suggested that this pressure natriuresis phenomenon plays an important physiological role in the maintenance of normal blood pressure (3) (4). A key element in their theory is that increments in blood pressure are dissipated by the volume loss caused by the pressure natriuresis and contrarily small degrees of hypotension result in sodium retention until normal blood pressure is restored. In essence, the pressure natriuretic phenomenon is important in both blood pressure control and volume homeostasis.

Guyton and colleagues further suggest that dampening of the pressure natriuresis phenomenon may be the cause of several forms of hypertension (3) (5). They postulate that in these disease states a higher pressure is required to excrete ingested sodium and systemic blood pressure sets at a higher value. This hypothesis has led to the study of relationships between arterial pressure and salt excretion in several models of experimental hypertension (6) (7), and led us to study if aberrations of the pressure natriuresis function (PNF) might be the cause of hypertension in Dahl rats.

Dahl rats are particularly suited to study the role of PNF in the development of hypertension. There are two strains of animals with opposite genetically controlled propensities for high blood pressure. Chronic excess salt feeding evokes hypertension in sensitive rats (S) while resistant animals (R) remain normotensive (8). Furthermore, there is data suggesting that renal factors are important in producing and maintaining hypertension in these strains. When kidneys from S animals are grafted into R rats, hypertension develops and contrarily, when R kidneys are given to S animals, blood pressure is reduced (9) (10).

We therefore designed protocols to detect if the cause of high blood pressure in S rats was due to a genetically determined flattening of the PNF. The effect of pressure on renal sodium handling was examined in both Dahl strains subjected to diet of either high or low sodium content, combining micropuncture and an isolated perfused kidney technique. By examining kidneys from S animals before and after the development of hypertension and comparing results to those of R rats, we observed that S Dahl rats display a normal PNF prior to the development of high blood pressure, but dampening of the PNF after the development of frank hypertension. Alteration in the PNF therefore cannot be implicated in the salt evoked hypertension in this strain. However, once blood pressure is elevated, it may be maintained by an altered pressure natriuresis function.

MATERIAL AND METHOD

Materials

Five groups of Dahl's S and R donor rats, described in table I, were fed either low or high sodium diets. Content of each diet was 0.02 and 8.02 g % respectively. Normotensive Wistar rats fed standard laboratory chow were used for the experiment validating the isolated kidney perfusion technique. Food was withdrawn the evening before each experiment but the animal had free access to water.

Isolated Kidney Perfusion Technique

a) Isolation of Donor Kidney

Isolated kidneys were perfused by a modification of the method of De Mello and Maack (11). The major alteration introduced by us was the use of a perfusion medium containing red blood cells. Rats, anesthetized by intraperitoneal injection of phenobarbital (10 mg/100 g body wt) (Nembutal, Abbott), were placed on a water thermoregulated dissection table. A tracheotomy was performed. The femoral artery cannulated (with PE 50 polyethylene tubing) and arterial blood pressure was monitored throughout the operation with a strain gauge (Endevco 8503-4), a Wheatstone bridge (Endevco 8630) and a graphical recorder (Hewlett Packard 7100 BM). Whenever pressure decreased more than 20 mmHg, all surgical manipulations were temporarily suspended and, if the decrement continued, 0.5 to 1 ml of isotonic saline was administered via the jugular vein. Unlike others (11), we did not canulate the vena cava noting that this procedure occasionally leads to transient but very pronounced drops in arterial pressure when the final ligature of the vena cava is tightened. The remainder of the surgical preparation of donor rat, the procedure of kidney isolation and the arrangement of the isolated kidney in a micropuncture cup were essentially that described by De Mello and Maack (11). The perfusion circuit (fig 1) was also similar to that described by these authors except that all the perfusion tubing were included in high section tubes for thermoregulation at 37.5 C° with circulatory water. This minimized the temperature changes of the perfusate during the recirculation, as it is required by the presence of living erythrocytes in the perfusate.

b) Control of Perfusion Pressure

Accurate monitoring of perfusion pressure (PP) within the renal artery is requisite for interpreting the results of these experiments. Perfusion pressure was monitored by controlling pressure above the renal canula and the renal

Table I - Groups of Dahl rats used in this study

n	Groups	Diet	Exposed to diet from :	Systolic blood pressure in vivo *
7	R ₀	OSD [‡]	weaning	121±5 mmHg
6	S ₀	OSD	weaning	123±4 mmHg
7	R ₇	HSD**	7 weeks	126±5 mmHg
5	S ₃	HSD	3 weeks	136±2 mmHg
6	S ₇	HSD	7 weeks	162±4 mmHg

* at time of kidney isolation

‡ low sodium diet

** high sodium diet

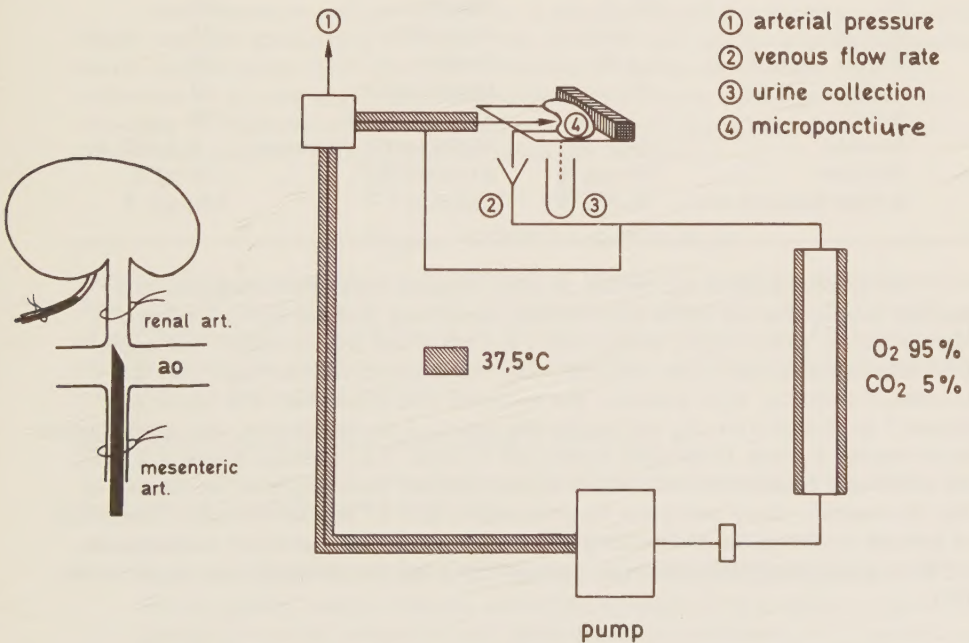


Figure 1 : Arrangement used for cannulation of renal artery and perfusion of isolated kidney preparation.

perfusion flow rate (PFR) *. Perfusion flow rate was measured by dividing the renal effluent with the aid of plexiglas tubing. The drops produced were counted by 2 electrodes and an electronical integrator. Pressure above the renal canula was continuously recorded by a strain gauge (Endevco 8503-4) and a Wheatstone bridge (Endevco 8630). Both PFR and pressure were simultaneously recorded on a 2 channels strip chart recorder (Hewlett Packard 7100 BM), calibrated in ml/min and mmHg.

c) Perfusion Medium

The composition of the perfusate is summarized in table II. This medium, except for the erythrocytes, is essentially that utilized by De Mello (11) with minor modifications (12).

Table II - Perfusate composition

Na	141 meq/l	Bovine erythrocytes	20 vol %
k	4.8	Methionine	7.5 mg %
Ca	5.9	Alanine	17.8
Mg	4.9	Glycine	15
Cl	115.7	Serine	21
H ₂ PO ₄	0.15	Arginine	21
SO ₄	4.8	Proline	23
HCO ₃	25	Isoleucine	13
HPO ₄	1.2	Aspartic acid	40
Acetate	10	Antidiuretic hormone	5.5 mU %
Glucose	100 mg %	Aldosterone	28 ng %
Bovine albumin fract. V	6 g %	Inulin	100 mg %

Ox blood, obtained by carotid section, immediately after slaughter in a packing house, was collected in a solution containing glucose H₂O : 13.2 mg/l, citrate tri-Na : 13.2 mg/l, citric acid : 4.4 mg/l and tetracycline : 150 mg/l (100 ml of solution per liter of blood). The blood was filtered through gaze and if clots or impurity were present, the material was discarded. The blood was stored 7 to 10 days at 4 C⁰, and on the day prior to an experiment, the erythrocytes were washed 4 times in isotonic saline and 2 times in a modified Krebs solution. An additional 2 washings in modified Krebs solution were repeated on the day of the experiment. Care was taken to centrifugate at 4 C⁰ and 2500 t/min. The pellet of washed erythrocytes had an hematocrit of about 70 %. A similar technique of erythrocytes preparation has been successfully used for isolated liver preparation (13).

* The canula (a modified Luer Lock needle : 2R2 N^o 12 Switzerland) offers an hydrolic resistance (R_h) which means that the perfusate pressure drops from the beginning of the needle to its tip. This pressure drop (ΔP) is proportional to PFR since :

$$\Delta P = \text{PFR} \times R_h$$

Canula ΔP , calculated for any observed PFR, must be subtracted from the enregistered pressure above the renal canula to allow the determination of PP.

Experimental Protocol

I) Wistar control kidneys : kidneys isolated from normotensive Wistar rats were perfused at a constant perfusion pressure of 125 mmHg. We noted that, during the first 20 minutes of perfusion, the pump had to be accelerated slightly in order to maintain the desired perfusion pressure. Once a steady state was achieved, 5 clearance collections were made over a period of 100 minutes.

II) Dahl experimental kidneys : in these experiments, perfusion pressure was increased from 105 mmHg * to 185 mmHg in 20 mmHg step. Once the steady state was achieved, a clearance period lasting 10 to 15 minutes was obtained. Then pressure was increased in stepwise fashion, while, after 5 to 8 minutes ** of equilibrium time, similar 10 to 15 minutes clearance periods were mesured. An aliquot of perfusate was obtained in the middle of each period. All urines were collected under oil in preweighted vials and the volumes were determined gravimetrically. Concentrations of inulin, sodium (UNa), potassium (UK) were determined on all samples by methods previously described (14) (15). These measurements permitted calculations of the glomerular filtration rate (GFR), urinary sodium reabsorption (% TNa), urinary sodium excretion (UNaV), urinary potassium excretion (UKV) and filtration fraction (FF) by standard formulae.

Microperfusion was performed in 12 kidneys during the perfusion procedure. Single nephron glomerular filtration rate (SNGFR) and tubular fluid over plasma inulin concentration ((TF/P)_{in}) were determined using conventional free flow collection at the end of proximal convolution (15). These values permitted calculations of fractional proximal fluid reabsorption (% T_{prox}) and end proximal tubule fluid delivery (FD_{prox}) by the following formulae :

$$\% T_{\text{prox}} = 1 - (1 / (TF/P)_{\text{in}})$$

$$FD_{\text{prox}} = \text{SNGFR} \times (1 / (TF/P)_{\text{in}})$$

Localisations were verified with injection of Microfil MV 122 (Canton Biomedical Product) and microdissection (16). Micropressure measurements using an adaptation of the technique of Wiederhielm et al (17) were determined in free flow proximal tubules, stop flow proximal tubules, free flow distal tubules and small peritubular capillaries 5 microns in diameter.

Measurement of Arterial Systolic Blood Pressure in Vivo

The determinations were performed under light ether anesthesia using the technique of Friedman and Freed (18).

-
- * When the donor rats were hypertensive, the experiments began at a PP of 145 mmHg, thus avoiding a too large pressure drop between in vivo systemic arterial pressure and initial perfusion pressure.
 - ** In order to verify if steady state conditions were maintained after increasing PP, clearance procedures were performed twice at 20 minutes intervals, once perfusion pressure reached 185 mmHg. Urinary sodium excretion amounted to 9.76 µeq/min/gr and 9.90 µeq/min/gr for the first test and 11.97 and 11.93 µeq/min/gr for the second test.

Statistics

The results are given as mean $\pm$ SE. The significance of the effect of increasing pressure among the different groups of kidneys was calculated using a variance analysis or the least squares method. Differences in means were checked using the Student t test.

RESULTS

I. Validation of the Isolated Kidney Perfusion Technique

In experiments designed to test the stability of our preparation, 8 kidneys, obtained from Wistar rats, were perfused at 125 mmHg for 100 minutes, a period of time exceeding that used when studying the Dahl rats. Results are summarized in fig 2, which illustrates the stability of GFR, FF, % TNa, UNaV and UKV during the 100 minutes of perfusion. GFR averaged 0.45 ml/min/gr during the first and second clearance periods and then slightly decreased to 0.38 ± 0.14 ml/min/gr ($p=NS$) at the end of the experiment. The filtration fraction (FF) amounted 4.5 ± 0.9 % during the first hour and 4.0 ± 0.7 % after 100 minutes of perfusion in contrast to the markedly lower values of FF reached in isolated kidney preparation perfused with acellular medium. This is due to the lower blood flow obtained in perfusing the isolated kidney with a cellular medium (12). Fractional sodium reabsorption averaged 98.5 during the first and second clearance periods and then decreased only to 97.7 ± 0.6 % ($p=NS$) in the last period. Thus, UNaV, at constant perfusion pressure, was under 1 μ eq/min/gr during the first hour and rose only to 1.33 ± 0.7 μ eq/min/gr after 100 minutes of perfusion ($p=NS$). These values were reached with a physiological albumine concentration of 6 gr % in the perfusate (19), in contrast to the concentration of 7.5 % necessary to reach comparable % TNa values when using an acellular perfusion medium (20). UKV decreased slightly with time from 0.55 ± 0.1 to 0.32 ± 0.08 μ eq/min/gr ($p=0.05$). Table III demonstrates the similarities of glomerular and tubular functions in isolated kidneys as compared to anesthetized kidneys in vivo. The distribution of intra-renal pressures in the isolated kidney preparation perfused with a cellular medium was determined. At a PP of 125 mmHg in the renal artery, the intra-renal pressures amounted to 47.8 ± 2.5 mmHg in the glomerular capillaries, 14.7 ± 1.0 in the proximal tubules, 8.0 ± 2 in the peritubular capillaries and 10.7 ± 0.9 mmHg in the distal tubules. These values were close to those described in vivo as shown in table III.

II. Studies on Dahl Rats

a) Effect of Sodium Intake on Systolic Blood Pressure (Table I and fig 3)

Rats from both the R and S strains remained normotensive when fed with a low sodium diet (121.0 ± 5 and 122.5 ± 4 mmHg in groups R_0 and S_0 respectively). However exposure of S animals to high sodium diet for 3 and 7 weeks provoked hypertension (HTA) : blood pressure averaged 136.0 ± 2 mmHg and 161.7 ± 3 mmHg in the S_3 and in the S_7 groups respectively. R rats, exposed to the same diet for 7 weeks, remained normotensive. Their blood pressure averaged 125.7 ± 5 mmHg.

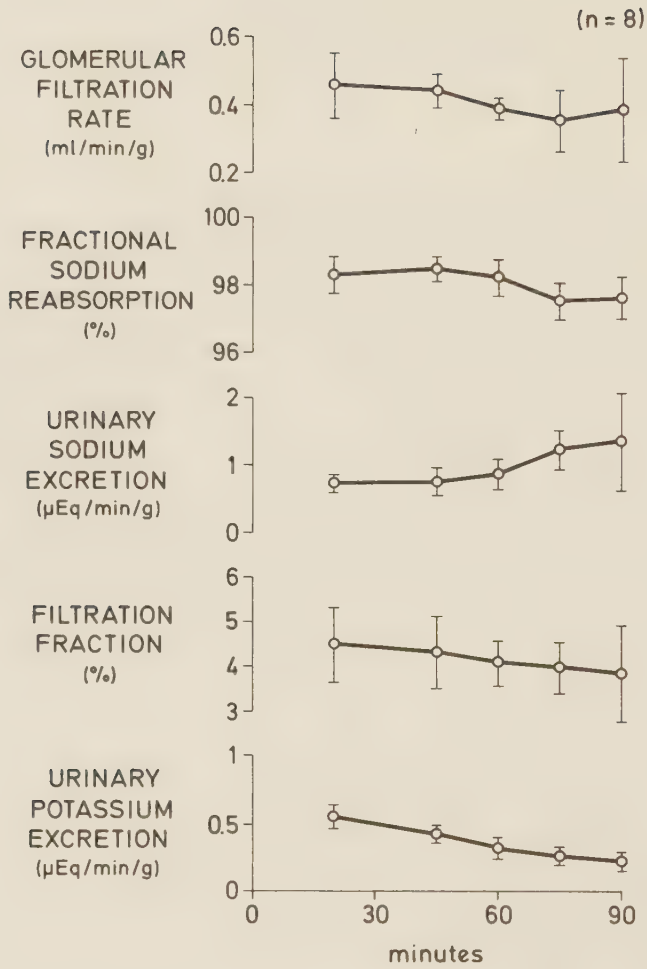


Figure 2 : Renal function in isolated kidneys from Wistar control rats.

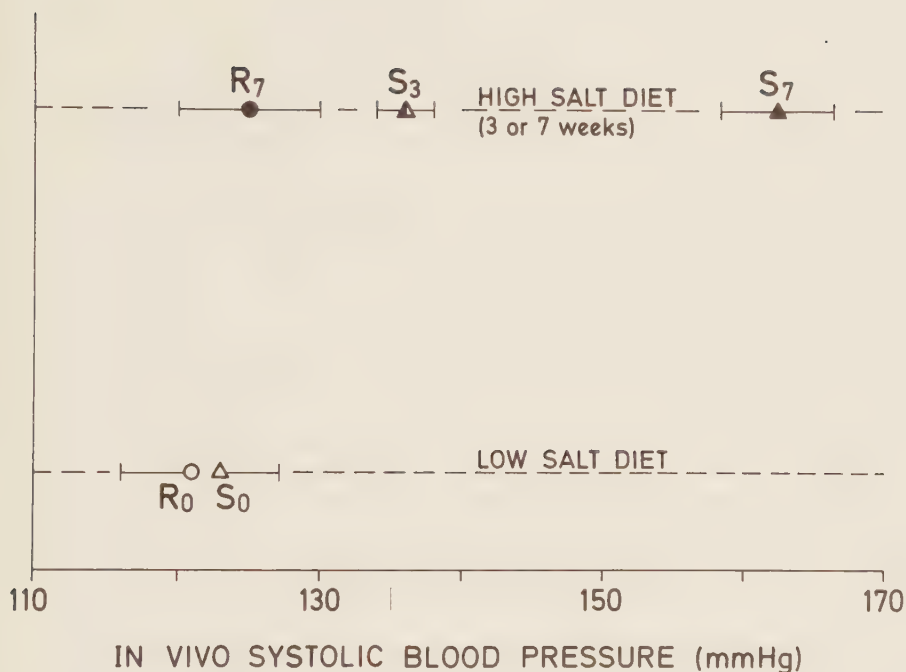


Figure 3 : Systolic blood pressure in the experimental groups of Dahl sensitive (S) and resistant (R) rats studied. The pressure were determined at time of kidney isolation. Donor rats were fed a low sodium diet in R_0 and S_0 groups or a high sodium diet for 3 and 7 weeks in S_3 and in S_7 and R_7 groups respectively. Each dots correspond to an equilibrium point as define in comments of figure 6 A.

b) Experiment on Isolated Kidneys

1. Animals fed low sodium diet. These studies were designed to detect any alterations in pressure natriuretic relationship in Dahl rats prior to their exposure to any hypertensiogenic stimuli. Fig 4 and table IV summarize effects on RPF, GFR, FF, % TNa, UNaV, UK and UKV in experiments where perfusion pressure was increased from 105 to 165 mmHg. GFR increased slightly with increased PP in S and R animals particularly after pressure had been increased above 145 mmHg ($p=NS$) and the increments were similar in both strains of animals (fig 4). Fractional sodium reabsorption decreased with increased PP in S_0 kidneys from 99.7 ± 0.2 % at 105 mmHg to 93.2 ± 1.1 % at 165 mmHg and in R_0 kidneys from 99.2 ± 0.2 % at 105 mmHg to 95.2 ± 0.9 % at 165 mmHg. This decrease was similar in both strains ($p=NS$). Baseline UNaV was similar in R_0

Table III - Renal function in control isolated kidneys as compared to in vivo conditions

	Isolated Wistar kidneys perfused with cellular medium *	Values of antidiuretic rats in vivo taken from ref. (21) (19) and (22)
Glomerular filtration rate	0.45 ml/min/g	0.50 ml/min/g ‡
Fractional sodium reabsorption	98.5 %	99 %
Urinary sodium excretion	0.80 µeq/min/g	0.53 µeq/min/g ‡
Glomerular capillaries pressures	47.8±2.5 mmHg	45.1±0.8 mmHg
Proximal tubules pressures	14.7±1.0 mmHg	13.4±0.3 mmHg
Distal tubules pressures	10.7±0.9 mmHg	7.4±0.4 mmHg
Peritubular capillaries pressures	8.0±2.0 mmHg	8.6±0.5 mmHg

* Mean values during first hour of perfusion

‡ The values were calculated from data in ref (21) and using the kidney weight versus body weight relationship found in our experimental rats which was 1 g kidney for a body weight of 220 g.

and in S_0 kidneys and excretion increased as PP was raised. The increment, too, was similar in both groups ($p=NS$): 0.5 ± 0.1 µeq/min/gr at 105 mmHg to 4.6 ± 0.8 µeq/min/gr at 165 mmHg in R_0 kidneys and 0.6 ± 0.2 µeq/min/gr at 105 mmHg to 5.7 ± 0.4 µeq/min/gr at 165 mmHg in S_0 kidneys.

Micropuncture of 7 end proximal convolutions in S_0 and 8 in R_0 kidneys were performed to determine the single nephron GFR ($SN\text{GFR}$), tubular fluid over plasma inulin ratio ($(TF/P)_{in}$), proximal tubular fluid reabsorption ($\%T_{prox}$) and fluid delivery at the end of the proximal tubules (FD_{prox}). None of these values correlated to the change in renal perfusion pressure from 105 to

145 mmHg. There was no significant difference of these values in the 2 groups of rats. SNGFR was 58.3 ± 8.4 and 50.5 ± 9.3 nl/min, $(TF/P)_{in} : 2.7 \pm 0.2$ and 2.6 ± 0.1 , $\% T_{prox} : 0.61 \pm 0.04$ and 0.62 ± 0.02 and $FD_{prox} : 23.0 \pm 3.6$ and 20.2 ± 4.7 nl/min in S_0 and R_0 kidneys respectively. Constancy in $\% T_{prox}$ and FD_{prox} indicate that pressure natriuresis might be due to a decreased sodium reabsorption located in the dilution segment of the nephron. These results are in accordance with the observation made by Di Bona et al in isolated dog kidney (23).

2. Animals fed high sodium diet. The response to increased PP in R_7 as well as S_3 and S_7 were designed to investigate the PNF in S strain animals after exposure to hypertensiogenic stimuli. Fig 4 and table IV summarize effects on RPF, GFR, FF, $\% TNa$, UNa , $UNaV$, UK and UKV in experiments where perfusion pressure was increased from 125 to 185 mmHg. With the rise in PP, GFR did not increase in S_7 kidneys as compared to the rise observed in S_3 and S_7 groups. It results that GFR was lower ($p < 0.01$) in kidneys from hypertensive S_7 donors than in kidneys of either S_3 or R_7 donors. At 185 mmHg, S_7 GFR amounted to 0.35 ± 0.05 ml/min/gr compared to 0.60 ± 0.05 and 0.73 ± 0.1 ml/min/gr in R_7 and S_3 kidneys. The decrements in $\% TNa$ during increments of PP were present in all groups and similar in S_3 and R_7 groups. At 145 mmHg, S_7 $\%TNa$ was comparable to others, thereafter, the response of S_7 kidneys seemed to diminish but by 185 mmHg, the difference was not significant. However, since GFR failed to rise in S_7 kidneys, $UNaV$ rose significantly less in S_7 kidneys than in the other two groups. S_7 $UNaV$ was 5.2 ± 0.9 μ eq/min/gr at 185 mmHg while R_7 and S_3 $UNaV$ were respectively 10.3 ± 0.4 and 12.3 ± 1.6 μ eq/min/gr.

3. Comparaison of results obtained between high and low sodium diet. The effect of sodium intake per se on kidneys isolated from normotensive donors may be seen by comparing results in R_0 and R_7 groups (fig 4 and table IV). GFR changes are similar in both R_0 and R_7 kidneys but $\% TNa$ is clearly greater ($p < 0.05$) in the kidneys from salt restricted animals and the difference is maintained at all PP levels.

4. Potassium excretion. Urinary potassium levels and excretion are summarized in fig 5 and in table IV. UKV was similar in R and S kidneys from normotensive animals at PP of 105 and 125 mmHg. However, with further increments of pressure, the increase in UKV was always greater in the S_0 strain ($p < 0.01$). After high salt feeding, the difference became more striking in the S_3 animals. However, the exaggerated kaliuresis is no longer present in the S_7 kidneys perhaps due to the relatively low GFR at high PP. S_0 and S_3 exaggerated kaliuresis might be related to higher levels of 18 OH DOC described in the sensitive strain (24) or might represent a more general pathology of potassium transport.

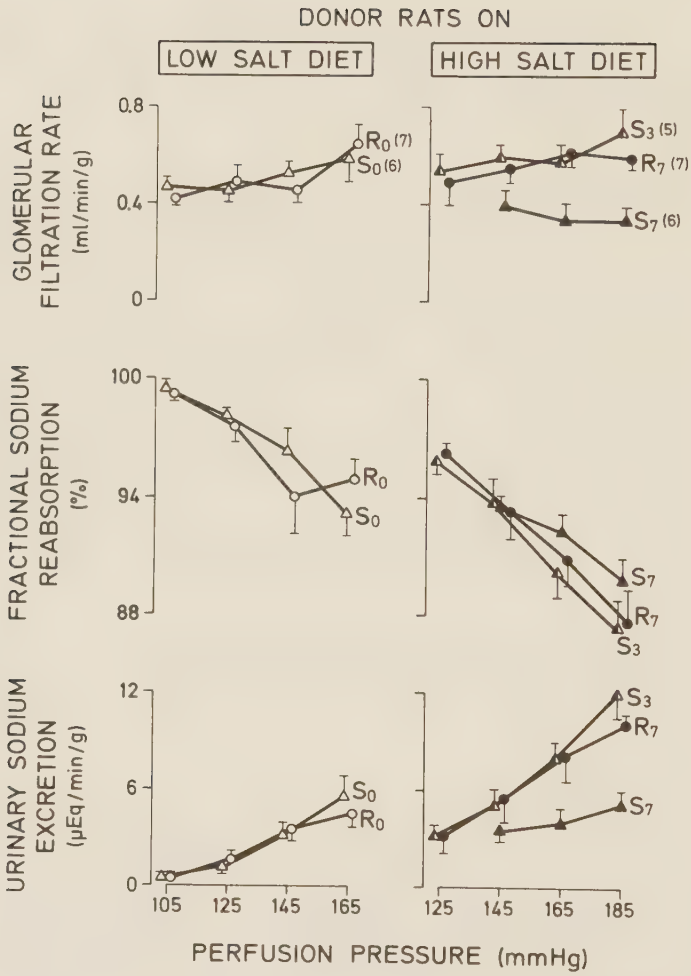


Figure 4 : Effect of varying the renal perfusion pressure on renal sodium transport in the 5 groups of Dahl rats studied.

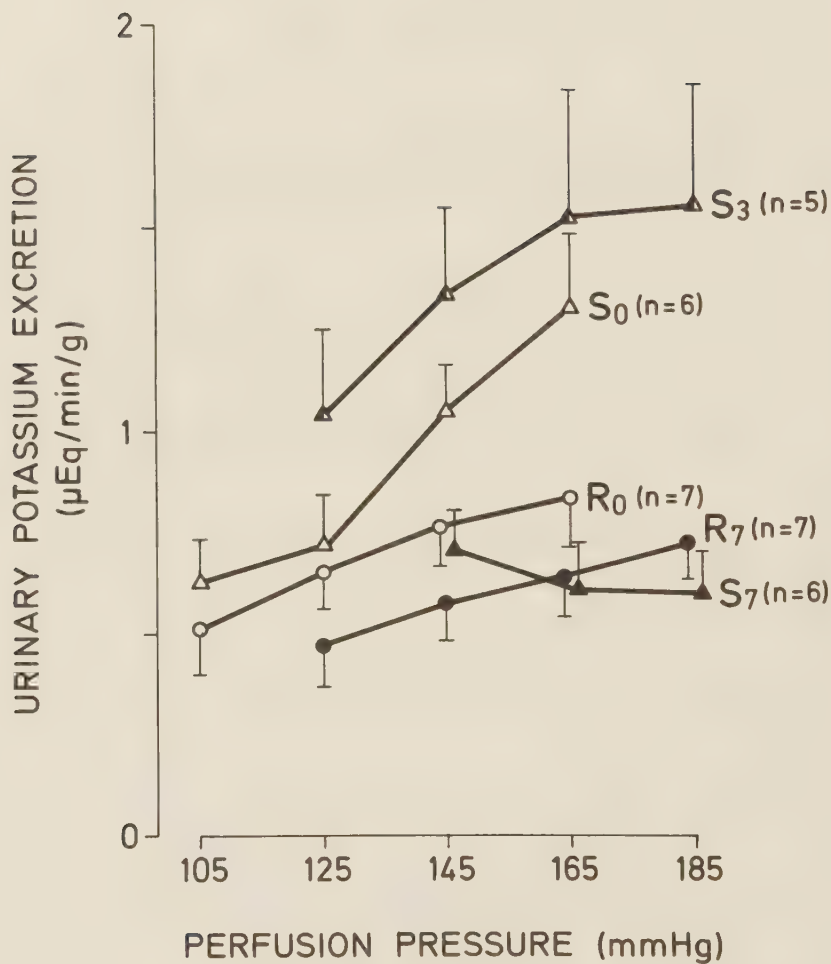


Figure 5 : Effect of varying the renal perfusion pressure on urine potassium excretion in the 5 groups of Dahl rats studied.

DISCUSSION

This study was designed to determine if an alteration of the renal pressure natriuretic function might be a cause of salt evoked hypertension in Dahl sensitive rats. The data demonstrate that there is no preset abnormality of this function when S rats are normotensive or at early stage of their hypertensive disease. However, once hypertension has been present for some time, there seems to be a resetting of the pressure natriuretic function, and this alteration may be a factor which sustains or aggravates high blood pressure in these rats.

Taking advantage of the salt sensitivity of Dahl rats, we varied both the sodium content and length of exposure to their diets, a maneuver which permitted us to study kidneys from sensitive normotensive animals, rats at early stage of their hypertension disease, hypertensive animals and appropriate controls (Table I and fig 3). In addition and in order to insure a more physiological perfusion, we modified existing methodology and were able to perfuse the experimental kidney with a cellular artificial solution, a technique differing from those of others who used acellular solutions or whole blood (containing uncontrolled humoral products) from donor animals (25) (26) (27) (28) (11) (6). We achieved a stable preparation whose function closely simulated that of *in vivo* antidiuretic rats, and then proceeded to study Dahl animals.

Since we have chosen to analyze our results in relation to the Guyton hypothesis (4), a review of his theory is in order. Guyton et al have stated that the slope characteristics of renal PNF *in vivo* defines an intercept (fig 6 A) at which an equilibrium between sodium intake and excretion is achieved. As a consequence, arterial blood pressure is stabilized to a pressure level determined by the location of the intercept. Guyton clearly stated that renal PNF, although describing a natriuretic effect of pressure acting through renal hydrolic mechanisms alone, is modified *in vivo* when sodium intake increases. In the latter situation, PNF is progressively changed, and in the schematic figure 6 B, curve number one becomes number five and equilibrium point D is achieved with very little change in arterial pressure (compare point D and E). In the absence of this adaptation, blood pressure would have risen to an hypertensive level define by equilibrium point H.

The data from resistant strain rats are in accordance with Guyton's views, and furthermore demonstrate that *in vitro* PNF is not merely a renal hydrolic mechanism, but was dependent on the metabolic and humoral status of the animal whose kidney was isolated. This is most evident when comparing results from R_0 and R_7 rats. *In vivo* blood pressure was similar regardless of the diet, but in the isolated kidney % TNa was lower, while UNaV greater in the R_7 animal, a difference which persisted at every perfusion level. As predicted by Guyton,

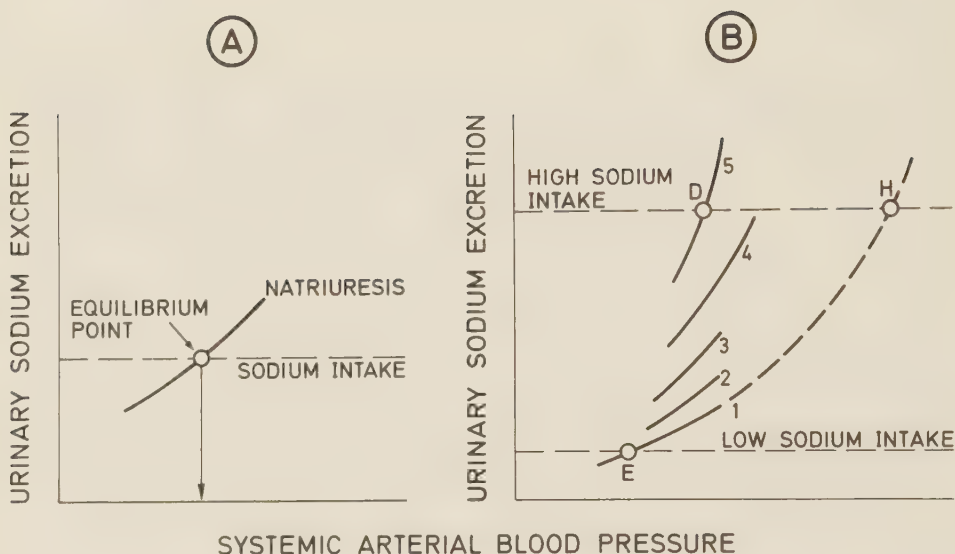


Figure 6 : Some aspect of Guyton theory on control mechanisms of arterial blood pressure. See explanations in text. (Adapted from ref (4)).

the PNF curve of R_7 is shifted to the left when compared to R_0 animals. If such in vitro data are applicable to the intact animal, PNF in the rat may be an important factor in maintaining blood pressure with increased salt intake.

Guyton has further suggested that certain forms of hypertension result from a dampening of the PNF (3) (5). In the present work, we used an isolated kidney preparation to ascertain whether the renal determinant of the slope of PNF might be responsible for the different location of the equilibrium points in the five groups of rats studied (fig 3). In such situations, high blood pressure would be produced by displacement to the right of PNF. It seemed plausible to us that this could well be the case in Dahl S rats since they remain normotensive on sodium restricted diets and become hypertensive with chronic excess salt feeding. We looked at the perfusion characteristics of the S_0 kidneys, for it is already here where a preset abnormality should be detected if our hypothesis was correct. However, the effect of pressure changes in R_0 and S_0 animals were identical. Furthermore when S_3 animals who are already at early stage of hypertension disease were examined, PNF was still unaltered in vitro since

we observed a shift to the left of the function similar to that of the resistant rats. Continued access to high sodium diets aggravates the hypertension in S rats and at seven weeks systolic blood pressure had increased to 160 mmHg. Now for the first time, alterations in pressure natriuretic relationship are present (fig 4). The curve is flattened in part due to an absence of rise in GFR, but also because % TNa has increased at high PP.

The reason why S₇ kidneys behave differently than all others is not apparent from our data, and we can only speculate on the cause. Extrusion of water across the vascular wall is known to occur in the presence of hypertension (29), and this may evoke a functional increase in the resistance of preglomerular arterioles. Also there are discrete histopathological changes in the kidneys of sensitive strain rats which as expected progress with sustained hypertension (30). Whatever the cause, the alteration in S₇ kidneys may be considered as an adaptation to hypertension and a resetting of the pressure natriuretic relationship which has occurred after the development of high blood pressure. The phenomenon of renal adaptation to increased blood pressure is a concept that is cited in the literature (6) (7) and our data are consistent with such a view. It should be noted that the secondary change in PNF may fix hypertension. In a teleological sense, hypertensive kidneys excrete sodium as resistant rats only at the cost of established hypertension.

Finally, although our results seems to eliminate alterations in PNF as the initiating cause of hypertension in Dahl sensitive rats, it should be noted that these data were obtained in *in vitro* circumstances and that *in vivo* experiments will be necessary to confirm our conclusion. Nevertheless the differences in R₀ and R₇ kidneys and the experiments in which potassium excretion was exaggerated in certain S suggest that our perfused kidneys retain some of the metabolic and humoral influences of their *in vivo* environment. Also our results do not eliminate other pathophysiological renal alterations as a cause of hypertension. It is known that transplantation of R kidneys into S animals ameliorates or avoid high blood pressure (9) (10). In this respect we have noted decrements in lipidic granules of the renal medulla in S animals * and others claim that these granules have an antihypertensive role (31). Furthermore we noted a potassium losing tendency in the S kidneys and others have demonstrate that the sensitive strain resists developing hypertension when chronic excess salt feeding is accompanied by a high potassium diet (32).

Further experiments should help to resolve these problems.

* Pricam et al ; manuscript in preparation

REFERENCES

1. Starling E.H., and E.B. Verney. 1924-25. The excretion of urine as studied in the isolated kidney. *Proc. Roy. Soc. (London). Series B* 97 : 321-363.
2. Selkurt E.E. 1951. Effect of pulse pressure and mean arterial pressure modification on renal hemodynamics and electrolyte and water excretion. *Circulation* 4 : 541-551.
3. Guyton A.C., T.G. Coleman, and H.J. Granger. 1972. *Circulation* : Overall regulation. *Ann. Rev. Physiol.* 34 : 13-46.
4. Guyton A.C., T.G. Coleman, A.W. Cowley, K.W. Scheel, R.D. Manning, and R.A. Norman. Arterial pressure regulation. Overriding dominance of the kidneys in long-term regulation and in hypertension. *Am. J. Med.* 52 : 584-594.
5. Coleman G.C., and A.C. Guyton. 1969. Hypertension caused by salt loading in the dog. 111. Onset transients of cardiac output and other circulatory variables. *Circulation res.* 25 : 153-160.
6. Tobian L., M.A. Johnson, J. Lange, and S. Magraw. 1975. Effect of varying perfusion pressure on the output of sodium and renin and the vascular resistance in kidneys of rats with "post-salt" hypertension and Kyoto spontaneous hypertension. *Circ. Research.* 36 : 37, suppl. 1 : 1-162-1-170.
7. Thompson J.M.A., and C.J. Dickinson. 1976. The relation between the excretion of sodium and water and the perfusion pressure in the isolated, blood-perfused, rabbit kidney, with special reference to changes occurring in clip-hypertension. *Clinical Science and Molecular Medicine* 50 : 223-236.
8. Knudsen K.D., L.K. Dahl, K. Thompson, J. Iwai, M. Heine, and G. Leitl. 1970. Effects of chronic excess salt ingestion. Inheritance of hypertension in the rat. *J. Exp. Med.* 132 : 976-1000.
9. Dahl L.K., M. Heine, and K. Thompson. 1974. Genetic influence of the kidneys on blood pressure. Evidence from chronic renal homografts in rats with opposite predispositions to hypertension. *Circ. Research* 34 : 94-101.
10. Dahl L.K., and M. Heine. 1975. Primary role of renal homografts in setting chronic blood pressure levels in rats. *Circ. Research* 36 : 692-696.

11. De Mello G., and T. Maack. 1976. Nephron function of the isolated perfused rat kidney. *Am. J. Physiol.* 231 : 1699-1707.
12. Girardin E., Y. Guyon, J. Caverzasio, and A. Grandchamp. Clearance study of isolated rat kidney perfused with a cellular solution. Effects of hematocrite, and liver function. *Kidney International* : submitted for publication.
13. Assimacopoulos-Jeannet F., J.H. Exton, and B. Jeanrenaud. 1973. Control of gluconeogenesis and glycogenolysis in perfused livers of normal mice. *Am. J. Physiol.* 225 : 25-31.
14. Vurek G.G., and S.E. Pegram. 1966. Fluorometric method for the determination of nanogram quantities of inulin. *Analytical Biochemistry* 16 : 409-419.
15. Diezi J., P. Michoud, A. Grandchamp, and G. Giebisch. 1976. Effects of nephrectomy on renal salt and water transport in the remaining kidney. *Kidney International* 10 : 450-462.
16. Cortell S. 1969. Silicone rubber for renal tubular injection. *J. Appl. Physiol.* 26 : 158-159.
17. Grandchamp A., and E.L. Boulpaep. 1972. Effect of intraluminal pressure on proximal tubular sodium reabsorption. A shrinking drop micropuncture study. *Yale J. Biol. and Med.* 45 : 275-288.
18. Friedman M., and S.C. Freed. 1949. Microphonic manometer for indirect determination of systolic blood pressure in the rat. *Proc. Soc. Exp. Biol. Med.* 70 : 670-672.
19. Daugharty T.M., I.F. Ueki, D.P. Nicholas, and B.M. Brenner. 1972. Comparative renal effects of isoncotic and colloid-free volume expansion in the rat. *Am. J. Physiol.* 222 : 225-235.
20. Bowman R.H., and T. Maack. 1974. Effect of albumin concentration and ADH on H₂O and electrolyte transport in the perfused rat kidney. *Am. J. Physiol.* 226 : 426-430.
21. Landwehr D.M., R.M. Klose, and G. Giebisch. 1967. Renal tubular sodium and water reabsorption in the isotonic sodium chloride-loaded rat. *Am. J. Physiol.* 212 : 1327-1333.
22. Brenner B.M., J.L. Troy, T.M. Daugharty, and W.M. Deen. 1972. Dynamics of glomerular ultrafiltration in the rat. 11. Plasma-flow dependence of GFR. *Am. J. Physiol.* 223 : 1184-1190.
23. Di Bona G.F., G.J. Kaloyanides, and R.D. Bastron. 1973. Effect of increased perfusion pressure on proximal tubular reabsorption in the isolated kidney. *Proc. Soc. Exptl. Biol. Med.* 143 : 830-834.
24. Rapp J.P., and L.K. Dahl. 1972. Mendelian inheritance of 18- and 11-steroid hydroxylase activities in the adrenals of rats genetically susceptible or resistant to hypertension. *Endocrinology* 90 : 1435-1446.

25. Nishiitsutsuji-uwu J.M., B.D. Ross, and H.A. Krebs. 1967. Metabolic activities of the isolated perfused rat kidney. *Biochem. J.* 103 : 852-862.
26. Little J.R., and J.J. Cohen. 1974. Effect of albumine concentration on function of isolated perfused rat kidney. *Am. J. Physiol.* 226 : 512-517.
27. Silva P., B.D. Ross, A.N. Charney, A. Besarab, and F.H. Epstein. 1975. Potassium transport by the isolated perfused kidney. *J. Clin. Invest.* 56 : 862-869.
28. Schurek H.J., J.P. Brecht, H. Lohfert, and K. Hierholzer. 1975. The basic requirements for the function of the isolated cell free perfused rat kidney. *Pflügers Arch.* 354 : 349-365.
29. Tobian L., Olson R., and G. Chesley. 1969. Water content of arteriolar wall in renovascular hypertension. *Am. J. Physiol.* 216 : 22-24.
30. Jaffé D., L.E. Sutherland, D.M. Barker, and L.K. Dahl. 1970. Effects of chronic excess salt ingestion. Morphologic findings in kidneys of rats with differing genetic susceptibilities to hypertension. *Arch. Path.* 90 : 1-16.
31. Muirhead E.E., G.B. Brown, G.S. Germain, and B.E. Leach. 1970. The renal medulla as an antihypertensive organ. *J. Lab. Clin. Med.* 76 : 641-651.
32. Dahl L.K., G. Leitzl, and M. Heine. 1972. Influence of dietary potassium and sodium/potassium molar ratios on the development of salt hypertension. *J. Exp. Med.* 136 : 318-330.

